AD_____

Award Number: **W81XWH-05-2-0027**

TITLE: IMPACT: Imaging and Molecular Markers for Patients with Lung Cancer: Approaches with Molecular Targets, Complementary/Innovative Treatments, and Therapeutic Modalities

PRINCIPAL INVESTIGATOR: George Simon, M.D., Principal Investigator

CONTRACTING ORGANIZATION:
The University of Texas M.D. Anderson Cancer Center
Houston, TX 77030

REPORT DATE: February 2015

TYPE OF REPORT: Annual

PREPARED FOR: U.S. Army Medical Research and Materiel Command
Fort Detrick, Maryland 21702-5012

REPORT DOCUMENTATION PAGE

Form Approved
OMB No. 0704-0188

1. REPORT DATE *(DD-MM-YYYY)*	2. REPORT TYPE	3. DATES COVERED *(From - To)*
February 2015	Annual	1 Feb 2014 - 31 Jan 2015

4. TITLE AND SUBTITLE

IMPACT: Imaging and Molecular Markers for Patients with Lung Cancer: Approaches with Molecular Targets, Complementary/ Innovative Treatments, and Therapeutic Modalities

5a. CONTRACT NUMBER
W81XWH-05-2-0027

5b. GRANT NUMBER

5c. PROGRAM ELEMENT NUMBER

6. AUTHOR(S)

George Simon, M.D.

email: gsimon@mdanderson.org

5d. PROJECT NUMBER

5e. TASK NUMBER

5f. WORK UNIT NUMBER

7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)

The University of Texas M.D. Anderson Cancer Center
Houston, TX 77030

Email: gsimon@mdanderson.org

8. PERFORMING ORGANIZATION REPORT NUMBER

9. SPONSORING / MONITORING AGENCY NAME(S) AND ADDRESS(ES)
U.S. Army Medical Research and Materiel Command
Fort Detrick, Maryland 21702-5012

10. SPONSOR/MONITOR'S ACRONYM(S)

11. SPONSOR/MONITOR'S REPORT NUMBER(S)

12. DISTRIBUTION / AVAILABILITY STATEMENT
Approved for public release; distribution unlimited

13. SUPPLEMENTARY NOTES

14. ABSTRACT

The projects in this proposal specifically target several signal transduction pathways known to be critical for NSCLC pathogenesis including the EGFR pathway and the more downstream ras/raf/Mek/ERK pathway. These projects combine targeted approaches using molecular and imaging techniques to validate activity against a target and monitor response using imaging modalities specific to the receptor using either small molecules or targeted peptide approaches.

15. SUBJECT TERMS
Lung cancer, molecular markers, molecular imaging, targeted therapy

16. SECURITY CLASSIFICATION OF: Unclassified			17. LIMITATION OF ABSTRACT	18. NUMBER OF PAGES	19a. NAME OF RESPONSIBLE PERSON
a. REPORT	b. ABSTRACT	c. THIS PAGE			USAMRMC
U	U	U	UU	18	19b. TELEPHONE NUMBER *(include area code)*

Standard Form 298 (Rev. 8-98)
Prescribed by ANSI Std. Z39.18

TABLE OF CONTENTS

IMPACT: Imaging and **M**olecular Markers for **P**atients with **L**ung **C**ancer: **A**pproaches with Molecular Targets, **C**omplementary, Innovative and **T**herapeutic Modalities

INTRODUCTION

Lung cancer is the most prevalent cancer worldwide and the leading cause of cancer-related mortality in both men and women in the United States. Conventional multimodality therapies (surgery, radiation and chemotherapy) have reached a therapeutic ceiling in improving the five-year overall survival rate of non-small cell lung cancer (NSCLC) patients, clinically in large part due to chemo- and radiation-resistant locoregional and metastatic spread but ultimately due to poor understanding of the disease and its resistance to the therapy.

Lung cancer is a heterogeneous disease, resulting from accumulated genetic abnormalities over years, which thus requires a coordinated attack in a truly integrated fashion on multiple altered signal pathways. Emerging targeted therapy aims to target key molecular abnormalities in cancer and has succeeded in some tumor types such as chronic myeloid leukemia (CML) (Druker et al., 2004; Druker and Sawyers et al., 2001; Druker and Talpaz et al., 2001), gastrointestinal stromal tumor (Demetri et al., 2002), colon cancer (Hurwitz et al., 2003), and breast cancer (Howell et al., 2005). Thus, the incorporation of targeted therapy into conventional treatments appears to be a new promising approach to treatment of lung cancer.

The program project IMPACT has proposed to integrate targeted therapy in the lung cancer research program when initial clinical results showed disappointing response rates and survival benefit of epidermal growth factor receptor (EGFR) inhibitor gefitinib (Iressa™) for non-selected lung cancer patients (Herbst et al., 2002, 2003, 2004; Herbst, 2004; Kris et al., 2003; Giaccone et al., 2004). It aims to validate molecular mechanisms of targeted agents alone and in combination with chemo- and/or radiation therapies in preclinical and clinical settings. It also aims to develop effective molecular imaging and cancer cell-targeted peptide-based delivery tools to help improve efficacy of the targeted agents. Specifically, our objectives are:

- To validate preclinically and clinically several key signaling pathways and their agents for therapeutic potentials alone or in combination with each other or with chemo and /or radiotherapy
- To explore applications of molecular imaging for targeted therapy and identify cancer cell-targeted peptides for systemic delivery of therapeutic and imaging agents
- To discover and evaluate new molecular abnormalities and therapeutic predictors in lung cancer
- To develop an educational program for teens and young adults for smoking risk and resultant lung cancer occurrence.

IMPACT is composed of 6 research projects, 1 Biostatistics Core, 1 Molecular Pathology Core, 1 Molecular Imaging Core, 2 career development projects, and 2 developmental research projects. We note that an additional no-cost extension for this grant has been approved to allow completion of the clinical activities proposed for Project 2 (the sole project remaining active) in this report.

Project 1: Targeting epidermal growth factor receptor signaling to enhance response of lung cancer to therapeutic radiation.

(PI and co-PI: Raymond E. Meyn, Ph.D., Ritsuko Komaki, M.D.)

In spite of significant technical advances including intensity-modulated radiation therapy (IMRT) and chemoradiation, locally advanced lung cancer continues to have a dismal prognosis as many patients' tumors appear to be resistant to radiation therapy. The molecular basis for radiation resistance is not fully understood, but tumor cells have an enhanced survival response that involves increased capacity for DNA repair and suppressed apoptosis. Both apoptosis propensity and DNA repair capacity are thought to be partly controlled by the upstream signal transduction pathways triggered by EGFR activation, which is constitutively activated in many NSCLCs, and its activation leads to a radiation-resistant phenotype. We hypothesize that the response of NSCLC to radiation can be improved through the use of inhibitors of EGFR signaling.

Aim 1 To test the combination of external beam radiation and the selective EGFR-tyrosine kinase inhibitor erlotinib (Tarceva) in locally advanced NSCLC.

Summary of Research Findings
This aim was completed as reported previously.

Aim 2 To test the hypothesis that activation of the EGFR pathway leads to radiation resistance in NSCLC cells due to an enhanced capacity for repairing DNA lesions.

Summary of Research Findings
This aim was completed as reported previously.

Aim 3 To test the hypothesis that clinically useful inhibitors of EGFR signaling abrogate DNA repair capacity, restore apoptotic response and radiosensitize NSCLC cells.

Summary of Research Findings
This aim was completed as reported previously.

Aim 4 To test the hypothesis that targeting both EGFR and its downstream signaling pathways will have at least an additive radiosensitizing effect on NSCLC.

Summary of Research Findings
This aim was completed as reported previously.

Aim 5 To test whether the strategies developed in Specific Aims 2-4 have efficacy in a xenograft tumor model.

Summary of Research Findings
This aim was completed as reported previously.

Project 2: Molecular Imaging of EGFR Expression and Activity in Targeting Therapy of Lung Cancer

(PI and co-PI: Juri Gelovani, M.D., Ph.D.; Roy Herbst, M.D., Ph.D.)

Aim 1 To synthesize novel pharmacokinetically optimized ^{124}I and ^{18}F-labeled IPQA derivatives for PET imaging of EGFR kinase activity and conduct *in vitro* radiotracer accumulation studies in tumor cells expressing different levels of EGFR activity.

Summary of Research Findings
This aim was completed and summarized in previous reports.

Aim 2 To assess the biodistribution (PK/PD) and tumor targeting by novel ^{124}I and ^{18}F-labeled EGFR kinase-specific IPQA derivatives using PET imaging in orthotopic mouse models of lung cancer and compare *in vivo* radiotracer uptake/retention with phospho-EGFR levels *in situ*.

Summary of Research Findings
This aim was completed as reported previously.

Aim 3 Using selected ^{124}I or ^{18}F-labeled IPQA derivative, to conduct pre-clinical studies in animals with orthotopic models of lung cancer xenografts with different levels of EGFR expression/activity, and to assess the value of PET imaging as the inclusion criterion for therapy by EGFR inhibitors, as well as for monitoring the efficacy of treatment with EGFR-targeted drugs.

Summary of Research Findings
This aim was completed as reported previously.

Aim 4 Perform pilot clinical PET imaging studies with the optimized ^{124}I or ^{18}F-labeled IPQA derivative under the RDRC guidelines in patients with NSCLC undergoing adjuvant therapy before tumor resection or biopsy. Compare PET image-based measures of EGFR activity with immunohistochemical measures of phospho-EGFR *in situ*.

Summary of Research Findings

Conduct of Protocol:
Patient screening was initiated following regulatory approvals as noted above. The patient population that is eligible for this trial includes patients with stage IV adenocarcinomas of the lung harboring *EGFR* mutations.

We are now routinely checking for *EGFR* mutations in all patients with stage IV adenocarcinoma of the lung seen in the clinic. We are currently identifying approximately 60 new stage IV patients with *EGFR* mutations a year. This figure does not include the patients who are identified elsewhere to be *EGFR*-mutation positive and then seek their care here. The majority of these newly diagnosed patients will be eligible for this clinical trial; thus, potential trial candidates will be drawn from this cohort of patients.

To further enhance accrual, we have modified the protocol to allow for patients who have previously been treated to enroll. Since the median overall survival of patients identified to have *EGFR* mutations is two years, this approach increases the patient pool from which patients can be drawn to approximately 150 patients. We are now maintaining a database of patients who are *EGFR*-positive to enable identification of potential study candidates to identify and consent patients quickly once this protocol is ready for accrual.

Progress in the conduct of the protocol includes the treatment of the first cohort (three patients) on protocol. All three doses were delivered within 10% of the requested dose (on the high side) as per standard nuclear pharmacy regulations, as noted in the table presented below.

Date	Activity Requested (mCi)	Calibration Time*	Activity Dispensed (mCi)	Dispensed Time**	Activity at Calibration Time (mCi)	Notes
8/28/2014	2	2:00 pm	2.265	2:00 pm	2.265	Delivered in three syringes
9/23/2014	2	2:00 pm	2.2	2:00 pm	2.2	
11/18/2014	2	1:15 pm	2.74	12:38 pm	2.17	

* Calibration Time: Requested time for injection by the imaging center
** Dispensed Time: Time the radiopharmacist draws and assays the dose

Differences were noted between the activity at calibration time and the administered activity due to minimal delays in injection time (decay), residual in the lines, and slight differences in the dose calibrator measurements between the cyclotron and the PET facilities. Specifically, residual activity in the patients was observed to be 0.2, 0.45, and 0.4 mCi, and the delays were in the range of 15, 30, and 15 mins.

Per protocol, a statistical analysis (see also Core B, "Biostatistics and Data Management Core," report) was performed to determine the dose level for the next cohort of patients, which was determined to be 3.78 mCi per patient. All required regulatory approvals have been obtained to move forward with patient screening and accrual to the next cohort of 6 patients (3 males and 3 females).

Key Research Accomplishments:

- Completed accrual of the first cohort of patients (3 females), with all three doses of this novel agent delivered within 10% of the requested dose (on the high side) as per standard nuclear pharmacy regulations.
- Completed the required statistical analysis to determine the dose level for the next cohort of patients (3.78 mCi per patient).
- Obtained all required regulatory approvals to move forward with patient screening and accrual to the next cohort of 6 patients (3 males and 3 females) upon approval of pending NCE request.

Conclusions:

The first cohort of three patients was completed and evaluated. Differences between the activity at calibration time and the administered activity were noted in all 3 patients; thus, drug administration will be carefully monitored to ensure acceptable doses per protocol.

Based on organ dosimetry the appropriate dose escalation for the next cohort of 6 patients was determined to be no more than 3.78 (+,- 10%) mCi per patient. All required approvals to move forward with accrual to the next cohort (IRB, IND, HRPO) have been obtained, and we will initiate patient screening immediately upon approval of the pending NCE request.

The planned number of patients for the trial is 15, and the completion of the trial is estimated to be within 12 months after its initiation. We are on target to enroll the next cohort of 6 patients in Q2 of this academic year. Analysis of patient safety and dose recommendations upon completion of that cohort will determine if a third cohort of 6 additional patients will be feasible. We are submitting a request for a no-cost extension to support completion of the 2nd cohort for this innovative trial and assessment of the feasibility to complete the trial per protocol.

Reportable Outcomes:

None to date.

Project 3: Targeted Peptide-based Systemic Delivery of Therapeutic and Imaging Agents to Lung Cancer

(PI and co-PI: Renata Pasqualini, Ph.D., Wadih Arap, M.D., Ph.D.)

The studies outlined in this proposal focus on the use of peptide sequences with selective lung tumor-targeting properties. We will seek to validate these probes as delivery vehicles in drug and gene-targeting approaches. This approach directly selects *in vivo* for circulating probes capable of preferential homing into tumors. The strategy will be to combine homing peptides in the context of phage as gene therapy vectors. Given that many of our peptides also target angiogenic vasculature in addition to tumor cells, these studies are likely to enhance the effectiveness of therapeutic apoptosis induction and imaging technology.

Aim 1 To select peptides targeting primary and metastatic tumors in lung cancer patients.

Summary of Research Findings
This aim was completed as reported previously.

Aim 2 To validate receptors for targeting human lung cancer.

Summary of Research Findings
This aim was completed as reported previously.

Aim 3 To design tools for molecular imaging of lung tumors.

Summary of Research Findings
This aim was completed as reported previously.

Project 4: Inhibition of bFGF Signaling for Lung Cancer Therapy

(PI: Reuben Lotan, Ph.D.)

The survival of lung cancer patients is poor because this cancer is diagnosed at advanced stages. Therefore, improvements in early detection through the identification of molecular markers for diagnosis and for intervention combined with targeted chemoprevention are urgently needed. While the molecular events involved in lung cancer pathogenesis are being unraveled by ongoing large scale genomics, proteomics, and metabolomics studies, it is already well recognized that proliferation-, survival- and angiogenesis- promoting signaling pathways are amplified in lung cancer. Among the angiogenesis signaling pathways, the basic fibroblast growth factor (bFGF) and its transmembrane tyrosine kinase receptors (FGFRs) are playing important roles in addition to the well-studied vascular endothelial growth factor (VEGF) and its receptors (VEGFRs). Both types of angiogenesis signaling pathways, the VEGF/VEGFR and the bFGF/FGFR, have been detected in NSCLC and associated with lung cancer development. However, most efforts in preclinical and clinical trials have been directed to the VEGF/VEGFR pathway.

We hypothesize that bFGF triggers signaling pathways that contribute to malignant progression of lung cancers by stimulating tumor cell and endothelial cell proliferation and survival and augmenting angiogenesis. Therefore, agents that intervene in this pathway may be useful for lung cancer therapy either alone or in combination with agents that target the VEGF/VEGFR signaling pathways and/or with cytotoxic agents. We will address the following specific aims in order to understand the mechanism(s) underlying the *in vitro* and *in vivo* effects of bFGF on lung cancer and endothelial cells and the ability of bFGF inhibitors to suppress the growth of NSCLC *in vitro* and *in vivo.*

Aim 1 Determine the effects of bFGF on *in vitro* growth, survival, motility, invasion and angiogenesis of NSCLC cells and endothelial cells.

Summary of Research Findings
This aim was completed and summarized in previous reports.

Aim 2 Evaluate the relative potency of several inhibitors of bFGF binding to receptor (i.e., TMPP and analogs) in inhibiting effects of bFGF detected in Specific Aim 1 and evaluate the effects of these inhibitors in combination with paclitaxel on *in vitro* growth and survival of tumor cells.

Summary of Research Findings
This aim was completed and summarized in previous reports.

Aim 3 Evaluate anti-tumor activity (growth inhibition, apoptosis, suppression of angiogenesis) of the most effective inhibitor identified in Specific Aim 2 when used alone and in combination with paclitaxel in an orthotopic lung cancer model using luciferase-expressing NSCLC cells for *in vivo* bioluminescence imaging of tumor growth and response to treatment.

Summary of Research Findings
This specific aim was abandoned as previously reported.

Reportable Outcomes

Publications Not Previously Reported: Saintigny, Pierre, Erminia Massarelli, Steven Lin, Young-Ho Ahn, Yulong Chen, Sangeeta Goswami, Baruch Erez et al. "CXCR2 expression in tumor cells is a poor prognostic factor and promotes invasion and metastasis in lung adenocarcinoma." *Cancer Research* 2013 73(2): 571-582. PMID: 23204236. PMCID:PMC3548940

Aim 4 To investigate the expression of bFGF signaling components (bFGF, FGFR-1, FGFR-2, heparan sulfate, syndecan-1, and FGFR-3) by IHC staining of tissue microarrays (TMAs), and correlate the expression of bFGF/bFGFRs between tumor and non-malignant epithelial cells with angiogenesis.

Summary of Research Findings
This aim was completed and summarized in previous reports.

Project 5: Targeting mTOR and Ras signaling pathways for lung cancer therapy

(Project Co-leaders: Shi-Yong Sun, Ph.D., Suresh Ramalingam, M.D.)

Aim 1 To determine whether an mTOR inhibitor inhibits the growth of human NSCLC cells via G1 growth arrest or induction of apoptosis, and to identify the molecular determinants of mTOR inhibitor sensitivity.

Summary of Research Findings
This aim was completed and summarized in previous reports.

Aim 2 To determine whether the effect of mTOR inhibitors on the growth of human NSCLC cells is enhanced in the presence of a PI3K inhibitor or a MAPK inhibitor.

Summary of Research Findings
This aim was completed and summarized in the previous annual report.

Aim 3 To evaluate the efficacies of the combinations of rapamycin with LY294002 or U0126 in nude mice models of lung cancer xenografts *in vivo*.

Summary of Research Findings
This aim was completed and summarized in previous reports.

Aim 4 To conduct a pilot clinical biochemical induction trial to investigate the effect of RAD001 in operable NSCLC patients and identify molecular determinants of RAD001 sensitivity and prognosis.

Summary of Research Findings
This aim was completed and summarized in the previous report

Project 6: Identification and Evaluation of Molecular Markers in Non-Small Cell Lung Cancer (NSCLC)

(PI and co-PI: Ralf Krahe, Ph.D., Li Mao, M.D)

A better understanding of the lung cancer biology and an identification of genes involved in tumor initiation, progression and metastasis are an important first step leading to the development of new prognostic markers and targets for therapy. In the same context, identification of reliable predictive markers for response or resistance to therapy in NSCLC patients is also desperately desired for optimal delivery of targeted therapy and/or standard chemotherapy. The proposed studies aim to identify the two types of markers that would eventually help develop smarter clinical trials, which will selectively recruit patients who are more likely to respond to one regimen over another and lead to improvement of overall therapeutic outcomes.

Aim 1 To expression profile by DNA microarray technology aerodigestive cancers - with primary focus on adenocarcinoma and squamous cell carcinoma (SCC) of the lung, and head and neck squamous cell carcinoma (HNSCC), including primary tumors and normal adjacent tissue, and (where available) metastatic lesions.

Summary of Research Findings
This aim was completed as reported previously.

Aim 2 To DNA profile the same samples by complementing DNA approaches to stratify RNA expression profiles on the basis of their corresponding DNA profiles.

Summary of Research Findings
This aim was completed as reported previously.

Aim 3 To evaluate the contribution of promoter hypermethylation and transcriptional inactivation of known cancer genes subject to epigenetic silencing to cancer phenotype.

Summary of Research Findings
This aim was completed as reported previously.

Aim 4 To determine protein signatures of treatments of erlotinib and other therapeutic agents, alone or in combination, in NSCLC and identify molecular predictors of response.

Summary of Research Findings
This aim was completed as reported previously.

Aim 5 To determine a clinical utility of the molecular predictors.

Summary of Research Findings
This aim was completed and summarized in previous reports.

Core B: Biostatistics & Data Management Core

(Core Director: J. Jack Lee, Ph.D.)

The Biostatistics and Data Management Core has continued to work with all IMPACT Projects in their research efforts, especially in the area of biostatistical support in clinical trial design, implementation, and analysis of experimental results. We also developed statistical methods to enhance the design and analysis pertinent to the lung cancer research.

Specific Aims:

1. To ensure that the results of all projects are based on well-designed experiments and are appropriately interpreted by providing experimental design; sample size estimates; power calculations; and integrated, comprehensive analysis for each basic science, pre-clinical, and clinical study.
2. To develop a data management system that integrates clinical, pathological, and basic science data while providing data integrity through process tracking and quality control.
3. To provide statistical and data management support for genomic and imaging studies including microarray, proteomics, and molecular targeted imaging.
4. To develop and adapt innovative statistical methods pertinent to biomarker-integrated translational lung cancer studies.
5. To produce statistical reports for all projects.
6. To collaborate and assist all project investigators with the publication of scientific results.

Summary of Research Findings and Key Accomplishments

For Project 2, "Molecular Imaging of EGFR Expression and Activity in Targeting Therapy of Lung Cancer," Core B worked with study investigators in the previously described revisions of the protocol "A phase I study of 18F-Fluoro-PEG6-IPQA as a PET Imaging Agent for Active/Mutant EGFR Expression in Tumors (2009-0832)," and completed the required statistical analysis of the first cohort accrued to the trial.

Statistical Analysis: Up to 15 patients will be imaged in this study using ^{18}F-PEG6-IPQA as a PET imaging agent. The maximum allowed single absorbed radiation doses for sensitive organs (whole body, gonads and red marrow) and non-sensitive organs are 3 and 5 rems, respectively. Absorbed dose estimates for 25 organs must be monitored. The study is designed with the intent to limit the probability that a patient exceeds the target dose (e.g., 3 or 5 rems depending on the organ) in any organ to be less than 0.10.

We proposed a three-stage design in which three female patients in the first stage and six patients (three females and three males) in each of the 2nd and 3rd stages are imaged, for a total of 15 patients. Since imaging quality is usually better in smaller patients, we stipulated that the first three patients were to be female and small in size, to maximize the likelihood of achieving useful data with the starting level of administered dose/activity.

Following the completion of each stage, the 90th percentile for the distribution of equivalent dose per unit administered activity (1 rem/MBq = 10 mSv/MBq) for the highest radiation dose administered will be estimated and used to determine the acceptable administered dose levels (or activities) for the next cohort. The initial administered dose level, determined as described below based on primate experiments, will be 70 MBq. Radiation absorbed dose will be estimated and monitored after each patient study and the administered dose will be recomputed if any patient exceeds the allowed single-dose limit in any organ

The 90th percentile of each organ's exposure per unit (mSv/MBq) and its standard deviation will be updated after observing data from each stage using the following formula:

$$UB_{ij} = \overline{x}_{ij} + t_{1-\alpha_i, n-1}.s_{ij}, i = 1,2,...,25, j = 1,2 \quad (1)$$

Where *i* denotes organ, *j* denotes dose level, $\overline{x}_{ij}$ is the observed mean exposure/dose (mSv/MBq) of the i^{th} organ at j^{th} dose, S_{ij} is the observed standard deviation of the i^{th} organ at j^{th} dose, *n* equals the number of patients treated at each dose level, α_i is the one-sided type I error rate spent on the i^{th} organ. The family-wise alpha level will be maintained at 0.10 level,

$$\alpha = \sum_{i=1}^{25} \alpha_i = 0.10, \quad (2)$$

but will be distributed unevenly among organs based on how close the organ dose is to its limit.

The 90th percentile of each organ's dose (rems) is calculated as UB_{ij} x $Dose_j$ /10 ($Dose_1$=70 MBq). If for any organ, the 90th percentile of dose exceeds its limit, then dose escalation will not occur and the trial will stop. If the 90th percentile of dose in each organ does not exceed its dose limit, then the next cohort of patients can be administered the same or a higher activity. The next administered dose will be determined such that the upper bound of absorbed dose to any given organ will not exceed its limit, i.e.,

$$Dose_{j+1} = \min\{Limit_i/UB_{ij}*10, \ MAX\}, i = 1,2,\ldots,25, j = 1,2 \quad (3)$$

where $Limit_i$ is the exposure limit (3 or 5 rems) for the i^{th} organ and MAX is the pre-specified maximum administered dose level, 370MBq.

Analysis Results. With the exposure data from first cohort of three female patients (exposure data shown as Total1, Total2 and Total3), we estimated the average and standard deviation of the exposure for each organ (unit mSv/MBq). Based on these estimates and the alpha spending level presented in the table (such that the family-wise alpha level is maintained at 0.10 level), the **recommended dose for the next level is 140 MBq** (3.78 mCi), based on equation (3). (See also Table, attached below.)

TargetOrgan	Total1	Total2	Total3	Limit	Ave(Dose1)	SD(Dose1)	Alpha	UB	Recommended Dose 2 (MBq)
Adrenals	0.0119	0.0115	0.0098	5	0.01107	0.001115	0.0002778	0.0584	370
Brain	0.004	0.0039	0.0042	5	0.00403	0.000119	0.0002778	0.0091	370
Breasts	0.0048	0.0048	0.0046	5	0.00473	0.000101	0.0002778	0.009	370
Gallbladder Wall	0.214	0.0946	0.0232	5	0.1106	0.096401	0.065	0.3512	142.39
LLI Wall	0.0337	0.0497	0.055	5	0.04613	0.011089	0.001	0.2937	170.23
Small Intestine	0.0863	0.136	0.15	5	0.1241	0.033476	0.01	0.3572	139.96
Stomach Wall	0.0115	0.0129	0.0127	5	0.01237	0.000757	0.0002778	0.0445	370
ULI Wall	0.0941	0.147	0.162	5	0.13437	0.035669	0.015	0.3356	148.97
Heart Wall	0.008	0.0078	0.007	5	0.00759	0.000533	0.0002778	0.0302	370
Kidneys	0.0331	0.0275	0.0241	5	0.02823	0.004545	0.0002778	0.221	226.28
Liver	0.0514	0.0474	0.026	5	0.0416	0.013657	0.001	0.3465	144.29
Lungs	0.007	0.0068	0.0062	5	0.00666	0.000448	0.0002778	0.0257	370
Muscle	0.0077	0.0083	0.0084	5	0.00814	0.000356	0.0002778	0.0232	370
Ovaries	0.0214	0.0291	0.0317	3	0.0274	0.005356	0.001	0.147	204.09
Pancreas	0.0135	0.0131	0.0114	5	0.01267	0.001115	0.0002778	0.06	370
Red Marrow	0.0083	0.0096	0.0098	3	0.00924	0.00079	0.0002778	0.0428	370
Osteogenic Cells	0.0093	0.0097	0.01	5	0.00966	0.000306	0.0002778	0.0226	370
Skin	0.0047	0.0049	0.005	5	0.00486	0.000116	0.0002778	0.0098	370
Testes									
Spleen	0.0146	0.0166	0.0135	5	0.0149	0.001572	0.0002778	0.0816	370
Thymus	0.0055	0.0054	0.0054	5	0.00544	0.000079	0.0002778	0.0088	370
Thyroid	0.0044	0.0043	0.0045	5	0.00442	0.000091	0.0002778	0.0083	370
Urinary Bladder Wall	0.0925	0.0674	0.0682	5	0.07603	0.014266	0.002	0.3009	166.16
Uterus	0.02	0.0245	0.0264	3	0.02363	0.003287	0.0002778	0.163	184.02
Total Body	0.0099	0.011	0.0108	3	0.01058	0.000569	0.0002778	0.0347	370
							Sum(Alpha)=0.1		

Conclusions
Core B continued to provide statistical support for Project 2, including the development of relevant statistical methodologies.

Core C: Pathology Core
(Director: Ignacio Wistuba, M.D.)

The IMPACT interdisciplinary research proposal for studying targeted therapy of lung cancers has required extensive histopathologic, IHC, and molecular studies of cell and tissues specimens, which have been assisted, coordinated or performed by the Pathology Core. One of the most important roles of the Pathology Core has been to provide professional technical services for proper procurement, storage and use of human and animal tissues, as well as technical assistance for IHC analysis. In addition, the Pathology Core has provided assistance for collection and evaluation of tissue specimens in IMPACT clinical trials in lung cancer patients.

Aim 1 Develop and maintain repository of tissue, cell and serum specimens from patients with lung neoplasia, as requested by the various component projects.

Summary of Research Findings
This aim was completed as previously reported.

Aim 2 Develop innovative tissue and cell reagents from lung cancer patients for the investigation and validation of the molecular endpoints relevant to each component project.

Summary of Research Findings
This aim was completed as previously reported.

Aim 3 Process human and animal cell and tissues for histopathological, immunohistochemical (IHC) and molecular analyses, including tissue microdissection, as required by each component project.

Summary of Research Findings
This aim was completed as previously reported.

Aim 4 Perform and evaluate IHC analysis in human and animal cell and tissue specimens, as required by the various component projects.

Summary of Research Findings
This aim was completed as previously reported.

DRP-1: Treatment of Malignant Pleural Effusion with ZD6474, a Novel VEGFR and EGFR TK Inhibitor
(PI and co-PI: Roy Herbst, M.D., Ph.D., Carlos Jimenez, M.D.)

Recurrent malignant pleural effusion is a debilitating clinical problem that requires palliation with repeated therapeutic thoracentesis or pleurodesis (Putnam, J Surg Clin North Am 2002). Malignant pleural effusions have been associated with high levels of VEGF. Treatment with a VEGFR tyrosine kinase inhibitor resulted in a decrease in the amount of pleural effusion in an animal model (Yano, CCR 2000). We hypothesize that malignant pleural effusion formation in cancer patients can be decreased with ZD6474 (AstraZeneca), a VEGFR and EGFR tyrosine kinase inhibitor.

Aim 1 To determine clinical effect of ZD6474.

Aim 2 To investigate biological correlates.

Aim 3 To investigate radiographic correlates.

Aim 4 To assess quality of life.

Summary of Research Findings
This study was completed as previously reported.

DRP-2: TALK - Teens and Young Adults Acquiring Lung Cancer Knowledge

(PI: Alexander V. Prokhorov, M. D., Ph.D.)

Ninety percent of lung cancer cases in adults are direct results of smoking. In children and young adults, tobacco use remains a major public health problem in spite of the recent declines in smoking prevalence among children and adolescents. Over the past 2-3 decades, numerous factors of smoking initiation among adolescents have been thoroughly investigated. A considerable volume of literature is currently available providing important clues with respect to designing tobacco prevention and cessation among youth.

Focusing on this major public health problem – tobacco use among young individuals and lack of in-depth knowledge of lung cancer issues – Project TALK (Teens and Young Adults Acquiring Lung Cancer Knowledge) was conceived and funded as a smoking cessation/prevention pilot project for culturally diverse high-risk young populations that include school drop-outs, economically disadvantaged, and underserved. Using modern technologies, the Departments of Behavioral Science and Thoracic/Head & Neck Medical Oncology have joined their efforts to conduct this developmental project under the leadership of Dr. Alexander V. Prokhorov. The project will assist in making major advances in lung cancer education and prevention among youth. Project TALK will produce a CD-ROM-based education/behavior change for teenagers and young adults (15-24 years of age).

Aim 1 **Develop intervention program.** Focus groups will be held with adolescents and young adults to ensure we are capturing the essence of the program, using the right messages, and employing the appealing video and animated characters. (Years 1-2)

Summary of Research Findings
This aim was completed and summarized in previous reports.

Aim 2 **Develop and beta-test CD-ROM.** This includes the design of the animation, illustrations, scripts and accompanying videos. (Years 1-2)

Summary of Research Findings
This aim was completed and summarized in previous reports.

Aim 3 **Implement program in agreed upon locations and recruit young adults to participate in the study.** (Years 3-4)

Summary of Research Findings
This aim was completed and summarized in previous reports.

Aim 4 **Collect and analyze data.** (Years 3-4)

Summary of Research Findings
This aim was completed and summarized in previous reports.

Career Developmental Project (CDP1): Identification of Membrane Proteins in Bronchial Epithelia Cells as Biomarkers of Early Detection for Lung Cancer

(PI: Ja Seok Peter Koo, Ph.D.)

Lung cancer is the leading cause of cancer deaths. Early detection of the malignant lesion leads to an improved 5-year survival rate after surgical resection. Therefore, advanced screening tools are needed urgently to detect lung cancer at an early stage to improve control of such deadly lung cancer.

Aim 1 **To isolate membrane proteins uniquely expressed on the surface of squamous metaplasia using organotypically cultured bronchial epithelial cells.**

Summary of Research Findings
This aim was completed as previously reported.

Aim 2 **To identify differentially represented proteins using proteomics.**

Summary of Research Findings
This aim was completed as previously reported.

Aim 3 **To verify the differentially represented proteins using PCR, Western blotting, and immunocytochemistry.**

Summary of Research Findings
This aim was completed as previously reported.

KEY RESEARCH ACCOMPLISHMENTS

Project 2: Molecular imaging of EGFR expression and activity in targeted therapy of lung cancer

Key Research Accomplishments:

- Completed accrual of the first cohort of patients (3 females), with all three doses of this novel agent delivered within 10% of the requested dose (on the high side) as per standard nuclear pharmacy regulations.
- Completed the required statistical analysis to determine the dose level for the next cohort of patients (3.78 mCi per patient).
- Obtained all required regulatory approvals to move forward with patient screening and accrual to the next cohort of 6 patients (3 males and 3 females) upon approval of pending NCE request.

Core B: Biostatistics & Data Management Core

For Project 2, "Molecular Imaging of EGFR Expression and Activity in Targeting Therapy of Lung Cancer," Core B worked with study investigators in the necessary revisions of the protocol "A phase I study of 18F-Fluoro-PEG6-IPQA as a PET Imaging Agent for Active/Mutant EGFR Expression in Tumors (2009-0832)," and completed the analysis of the first cohort accrued to the trial.

REPORTABLE OUTCOMES

None.

CONCLUSIONS

Project 2: The first cohort of three patients was completed and evaluated. Differences between the activity at calibration time and the administered activity were noted in all 3 patients; thus, drug administration will be carefully monitored to ensure acceptable doses per protocol.

Based on organ dosimetry the appropriate dose escalation for the next cohort of 6 patients was determined to be no more than 3.78 (+,- 10%) mCi per patient. All required approvals to move forward with accrual to the next cohort (IRB, IND, HRPO) have been obtained, and we will initiate patient screening immediately upon approval of the pending NCE.

The planned number of patients for the trial is 15, and the completion of the trial is estimated to be within 12 months after its initiation. We are on target to enroll the next cohort of 6 patients in Q2 of this academic year. Analysis of patient safety and dose recommendations upon completion of that cohort will determine if a third cohort of 6 additional patients will be feasible. We are submitting a request for a no-cost extension to support completion of the 2nd cohort for this innovative trial and assessment of the feasibility to complete the trial per protocol.

Biostatistics Core: Core B continued to provide statistical support for Project 2, including the development of relevant statistical methodologies, and analysis of the first cohort of the clinical trial.